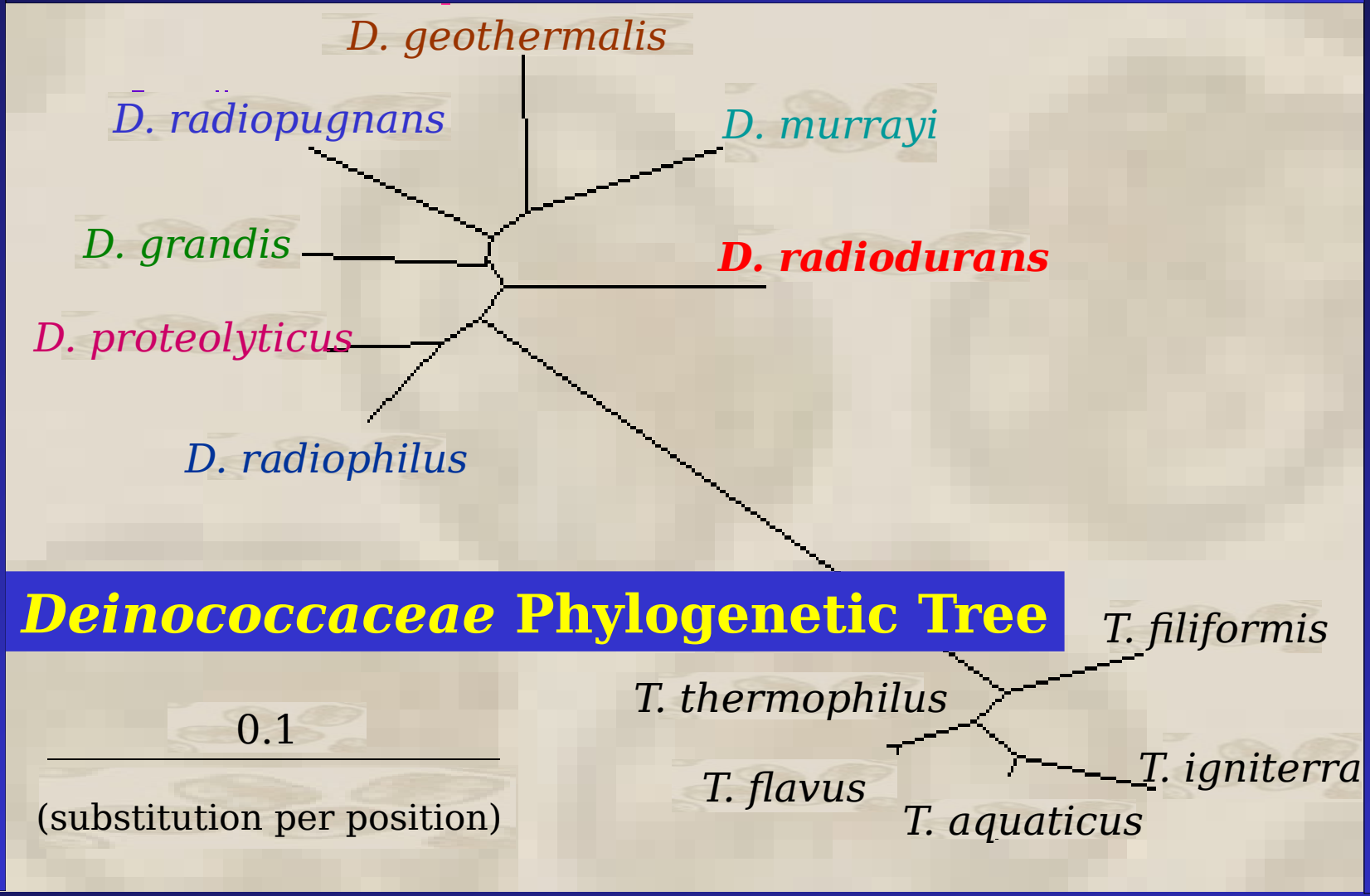
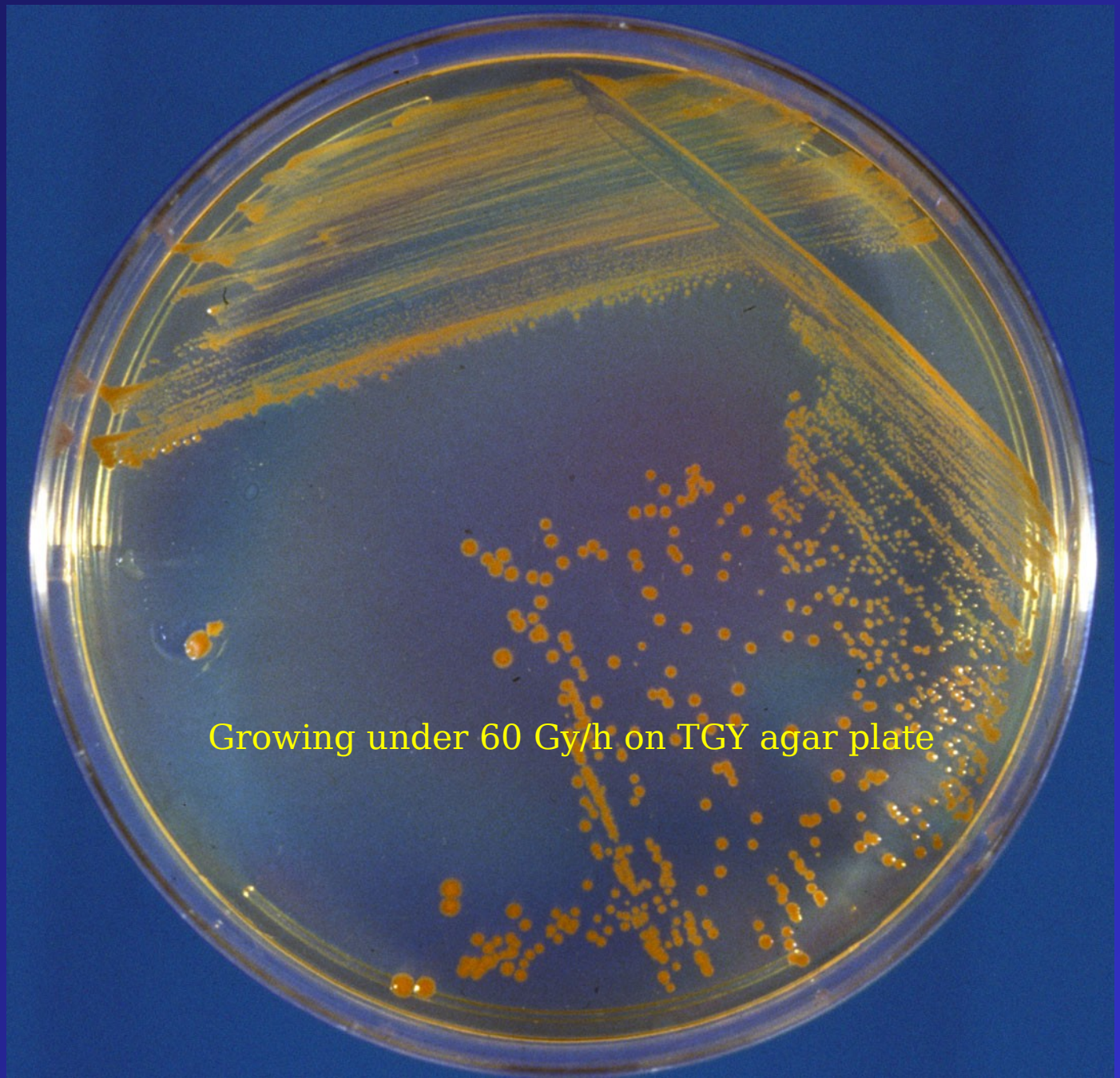
Deinococcus radiodurans - a radiation-resistant bacterium



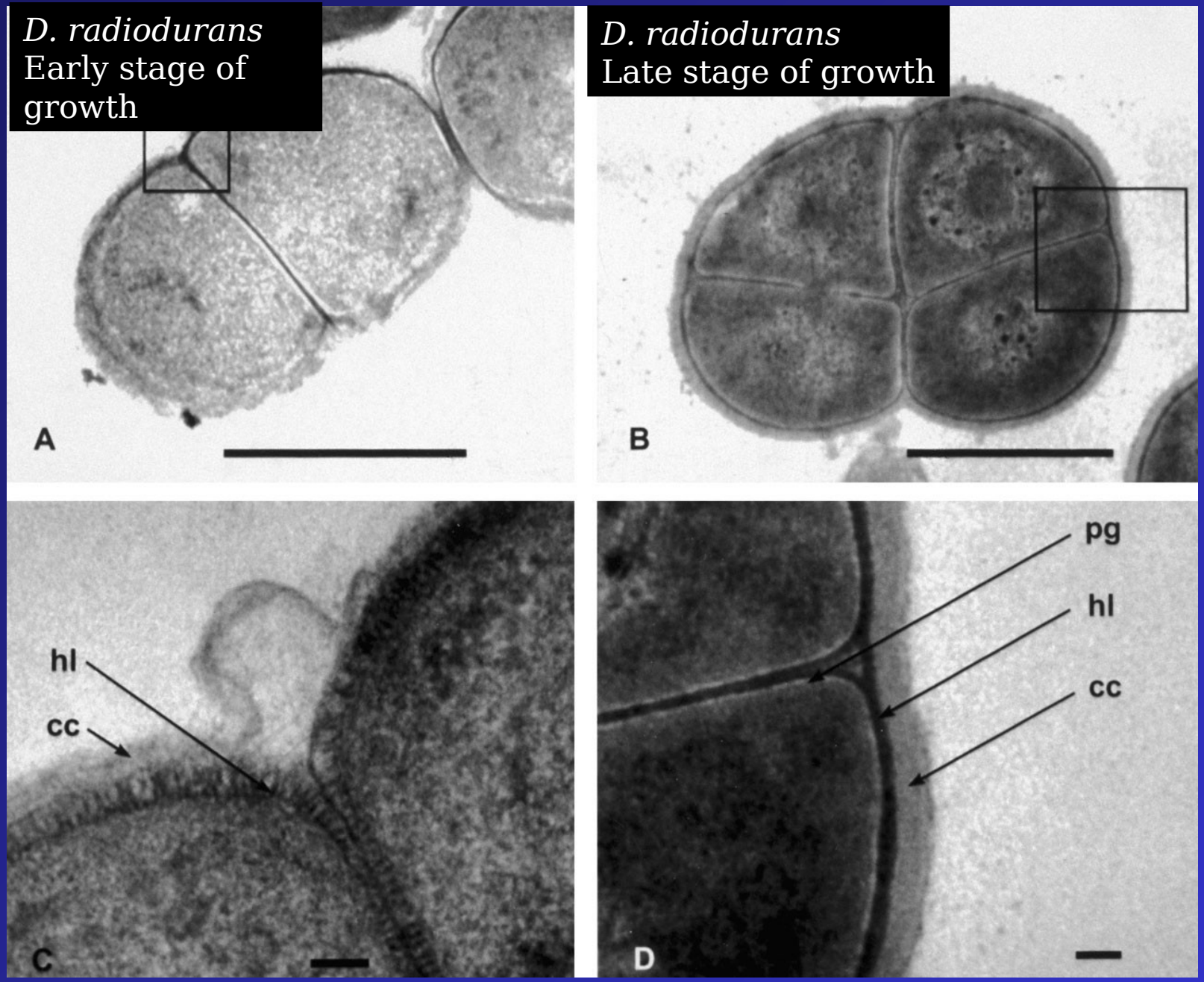
Neighbor-Joining unrooted phylogenetic tree was built using the PHYLIP program on th
rRNA sequences. Five *Thermus* species (*T. thermophilus, T. flavus, T. aquaticus, T. ignite*
are included as an outgroup. The *Deinococcus/Thermus* group is deeply branched in bac
trees with putative relationships with the actinobacteria/cyano-bacteria branch.The tree

be found elsewhere (see for example, Wolf et al., *BMC Evol. Biol*. 2001, Oct 20;1(1):8).

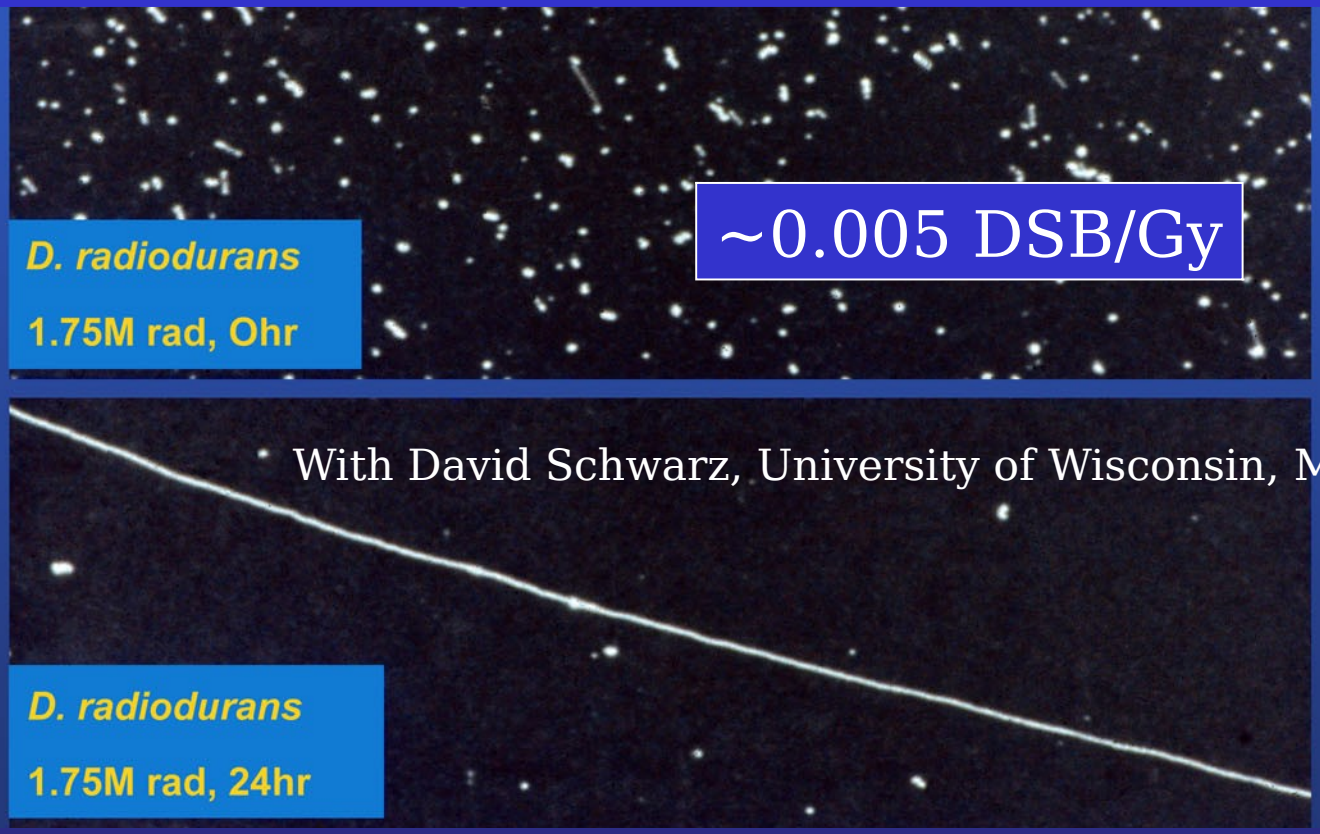
D. radiodurans



The Structure of *D. radiodurans*



D. radiodurans: The Ultimate DNA Assembly Machine

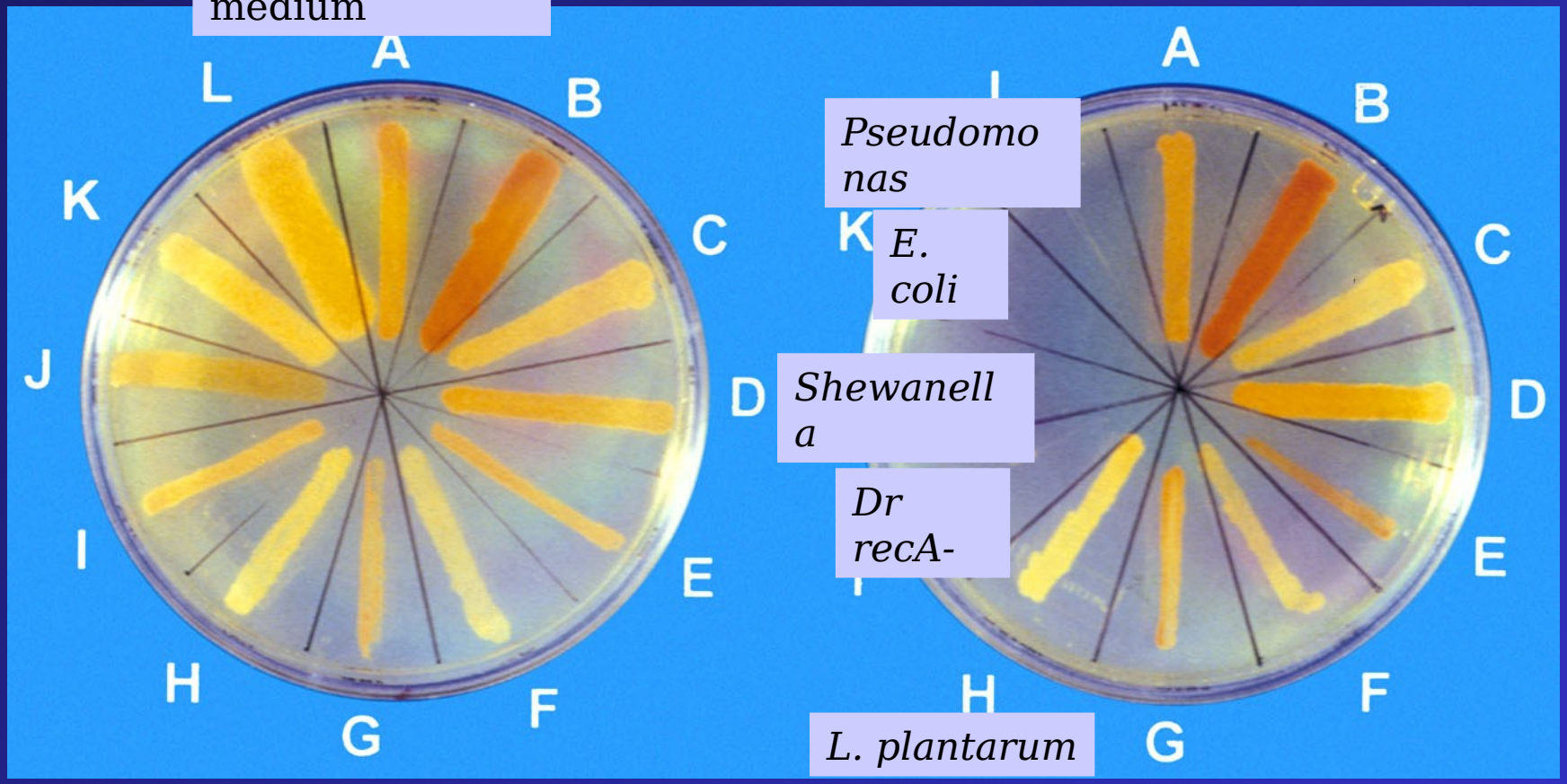


With David Schwarz, University of Wisconsin, Madison

Resistance to Chronic Irradiation

Control rich medium

+60 Gy/Hour



Pseudomonas

E. coli

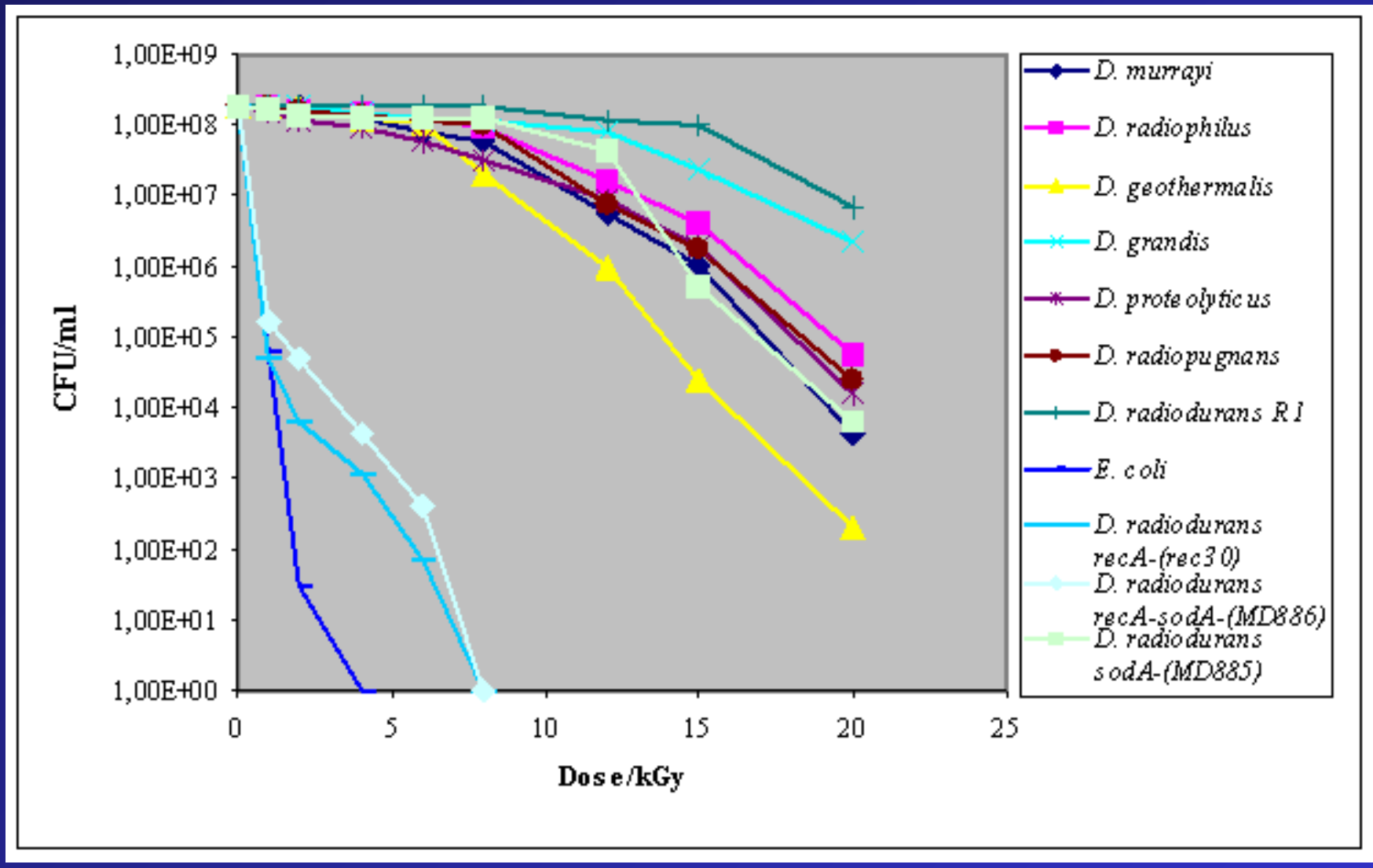
Shewanella

Dr recA-

L. plantarum

A-G, Deinococcaceae

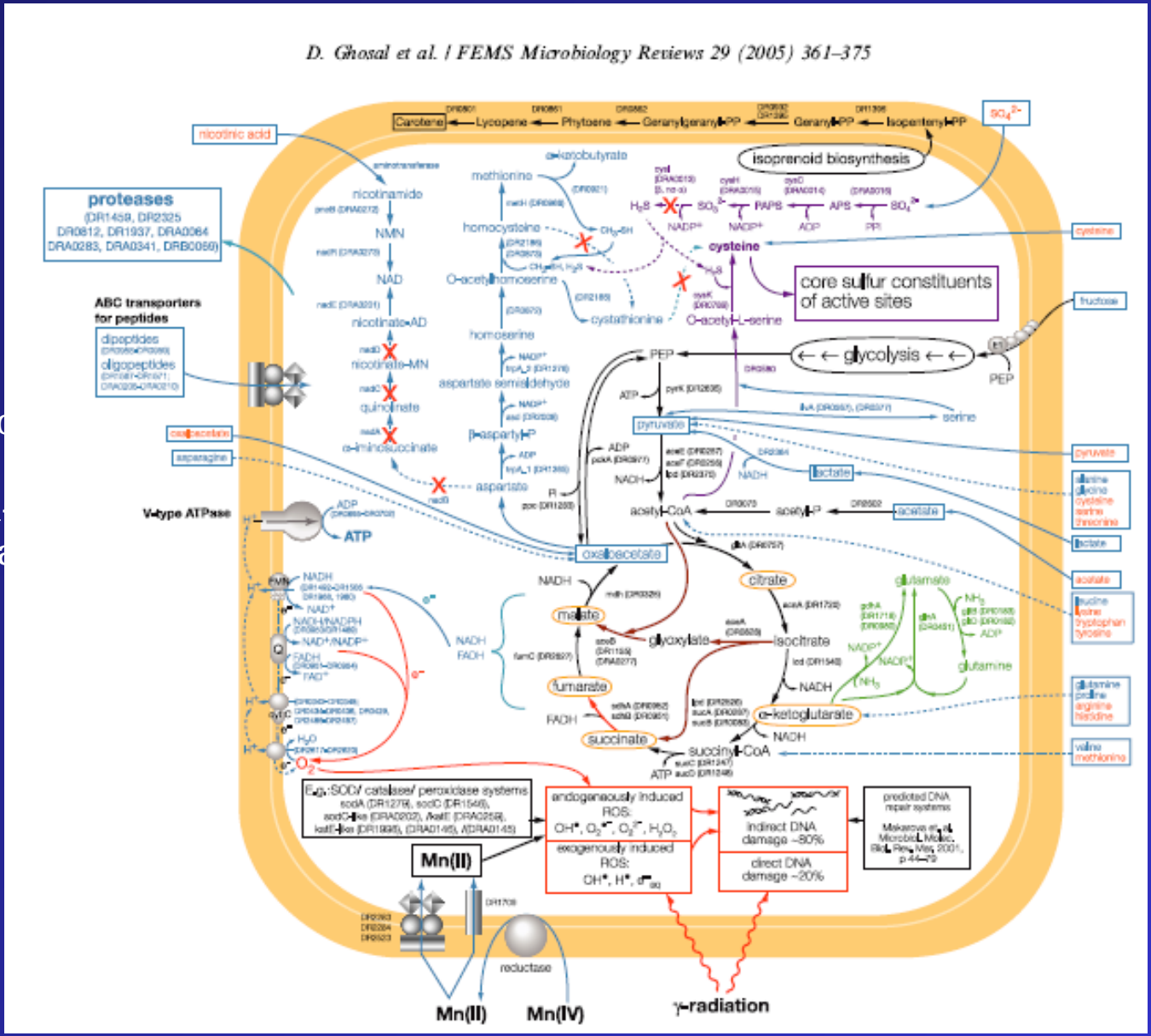
Resistance to Acute Irradiation



Predicted *Deinococcus* Metabolic Pathways

Red crosses indicate metabolic blocks which are predicted to facilitate the accumulation of metabolites.

Small-metabolite Mn complexes protect proteins from oxidation.



Statistical evaluation of survival of individual cells within a mixed population of tetracocci and diplococci

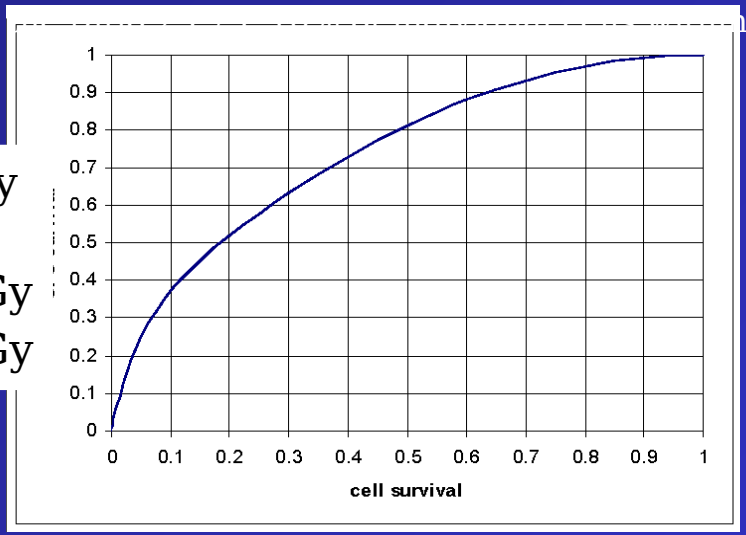
For the survival curve of *D. radiodurans* (OD_{600} 0.9; D_{37} 12 kGy) presented in Fig. 2, the cell-grouping was ~75% diplococci and ~25% tetracocci (Fig. 1a, and additional images at http://131.158.180.98/~alex/).

Under the assumptions that the survival of cells, constituting the CFU, is independent from each other, that survival of a single cell is enough to unsure the survival of a CFU, and that the relative frequency of a k-cell CFU is known to be f_k ($\Sigma f_k = 1$), the relationship between the individual cell survival and the CFU survival follows the equation:

$$p(x) = \Sigma f_k p_k(x) = \Sigma f_k (1 - [1 - p_1(x)]^k)$$

where, for a radiation dose x, $p(x)$ is the survival probability for an arbitrary CFU in the mixture, $p_k(x)$ is the survival probability for a k-cell CFU and $p_1(x)$ is the survival probability for an individual cell.

For a culture with a D_{37} value of 12 kGy and consisting of tetracocci and diplococci, in [illegible] the population will survive (D_{10}).



D. radiodurans D_{10} = 12 kGy

E. coli D_{10} = 0.75 kGy

S. oneidensis D_{10} = 0.1 kGy

The lack of a clearly identifiable unique DNA repair system in *D. radiodurans* has given rise to three competing views of the mechanisms responsible for its extraordinary survival

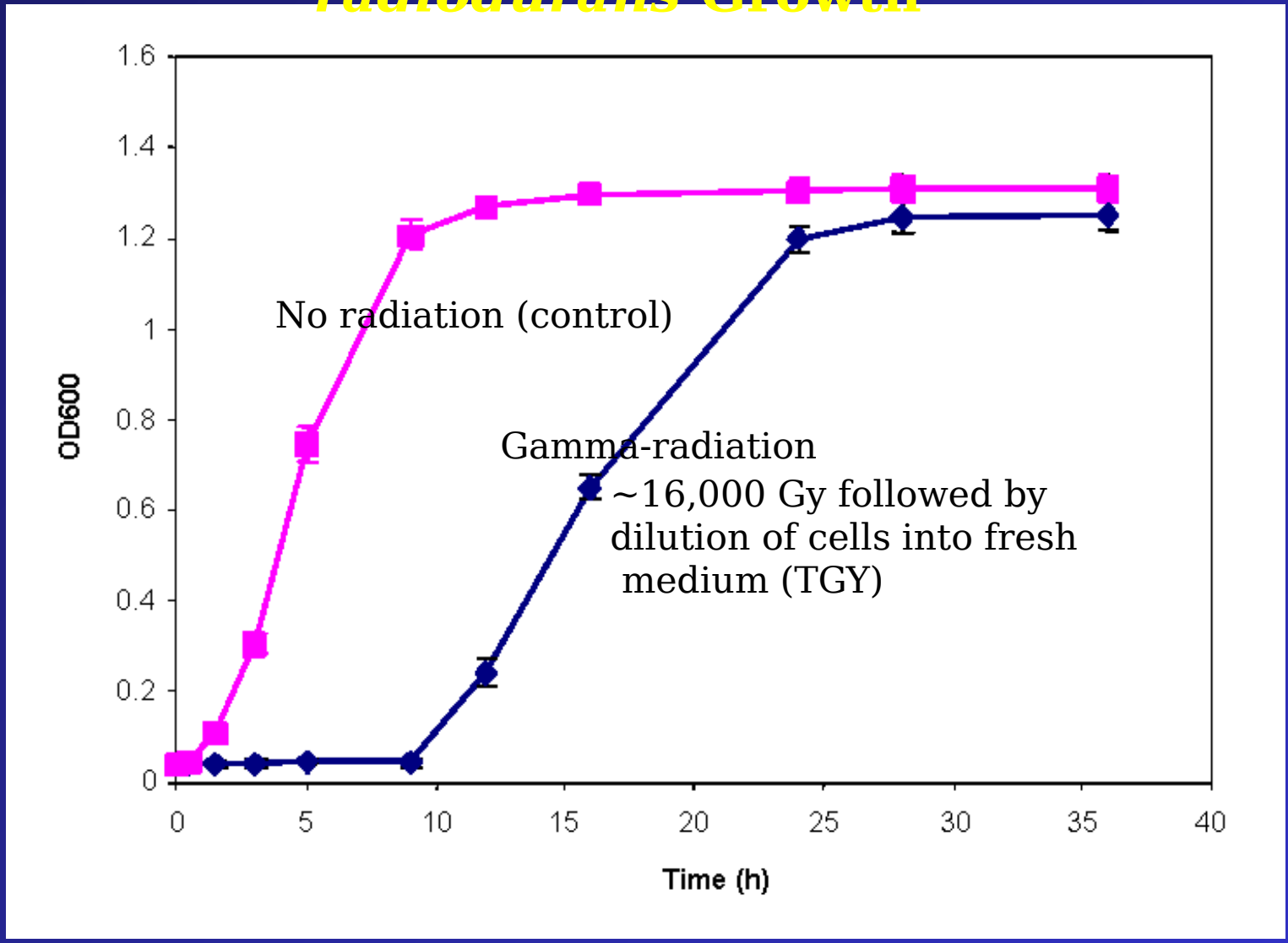
(i) there are novel repair functions encoded among hypothetical genes predicted by genomic annotation

(ii) *D. radiodurans* uses conventional DNA repair pathways, but with greater efficiency than other bacteria; or

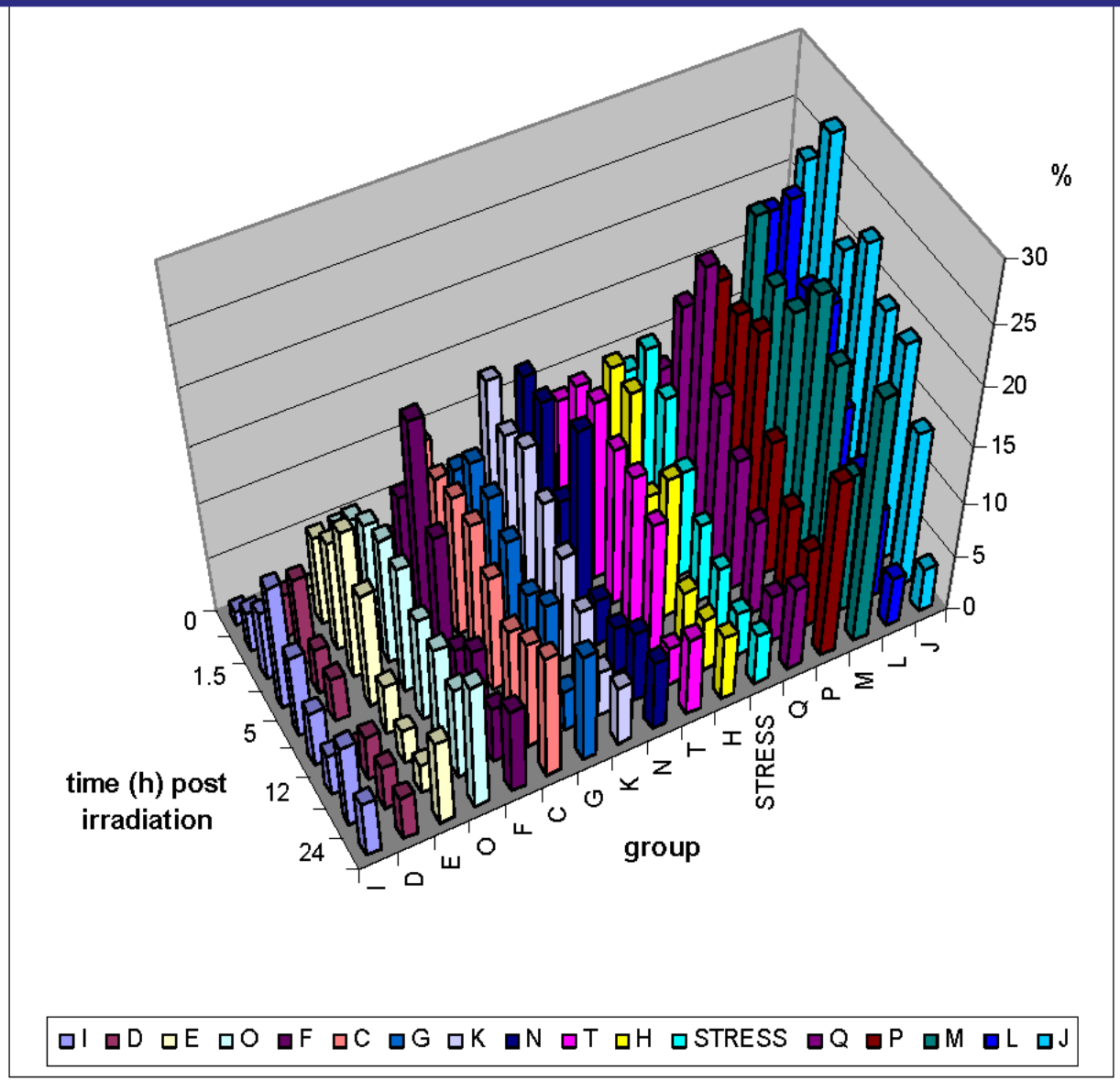
(iii) *D. radiodurans* contains potent Mn-dependent antioxidant complexes which prevent protein oxidation during irradiation, desiccation and other oxidative stress –generating conditions.

For a recent review: *Nature Reviews Microbiology* 7, 237-245 (2009).

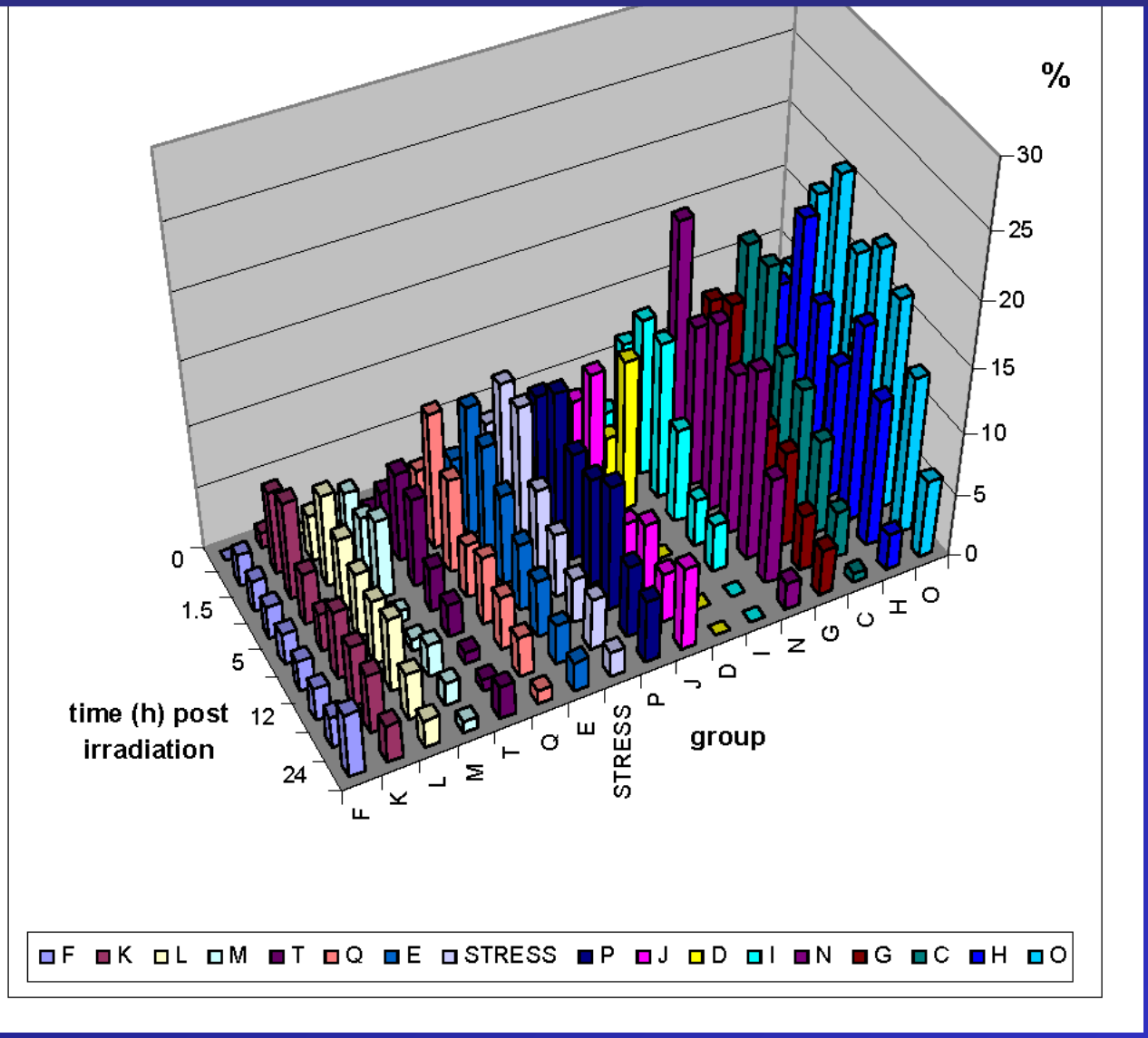
Effect of High-Dose Acute Irradiation on *D. radiodurans* Growth



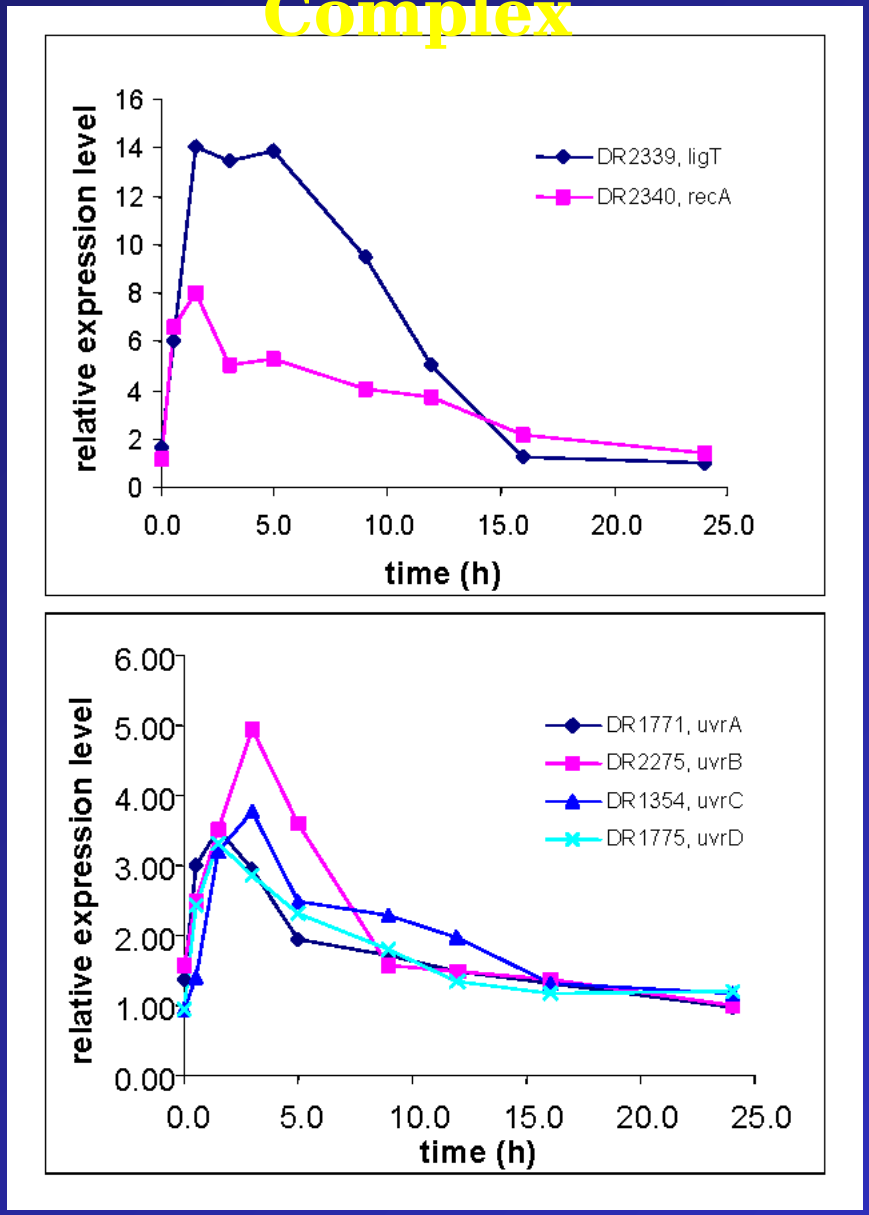
Up-Regulated Genes in Different Functional Gr



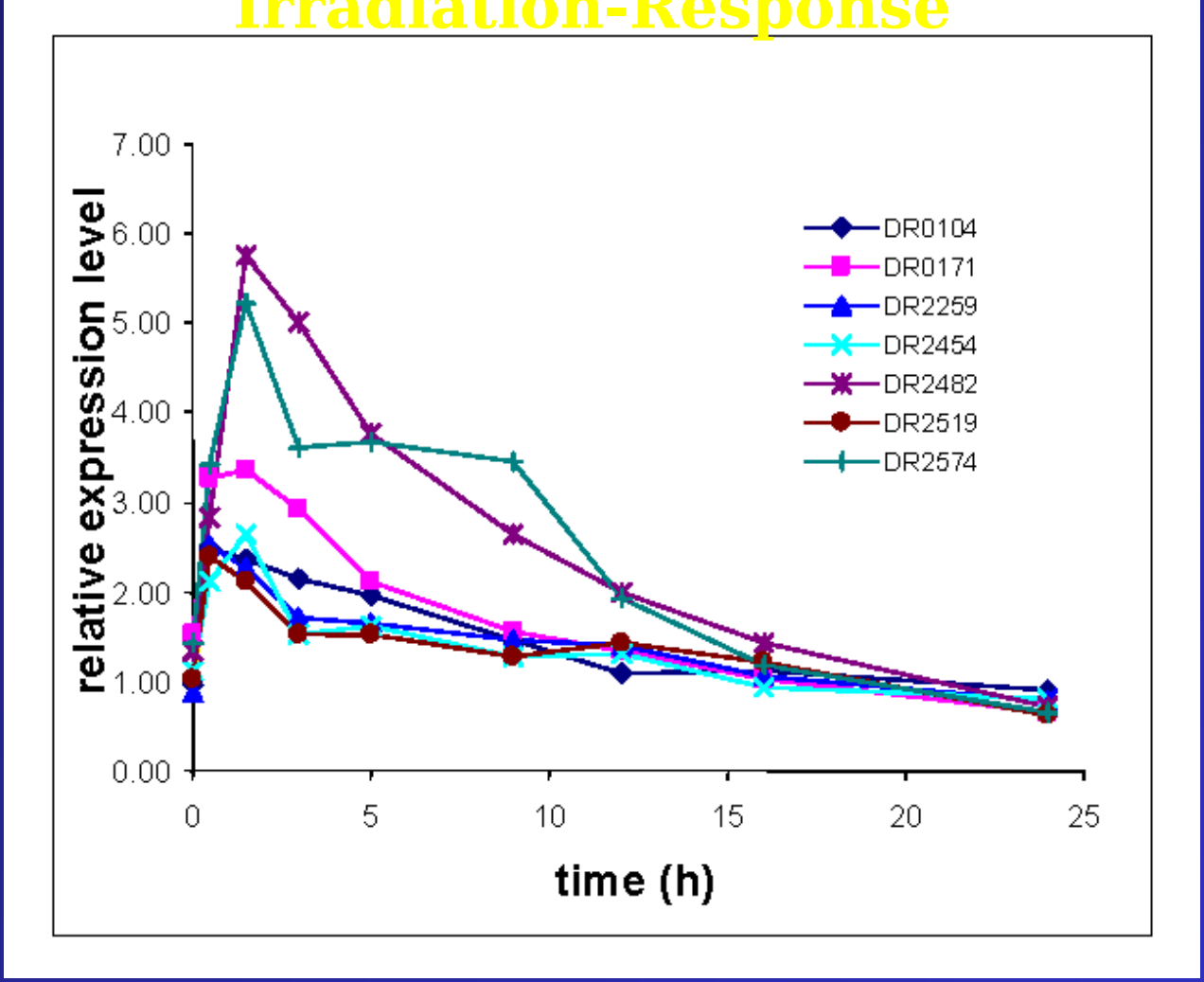
Down-Regulated Genes in Different Functional Groups



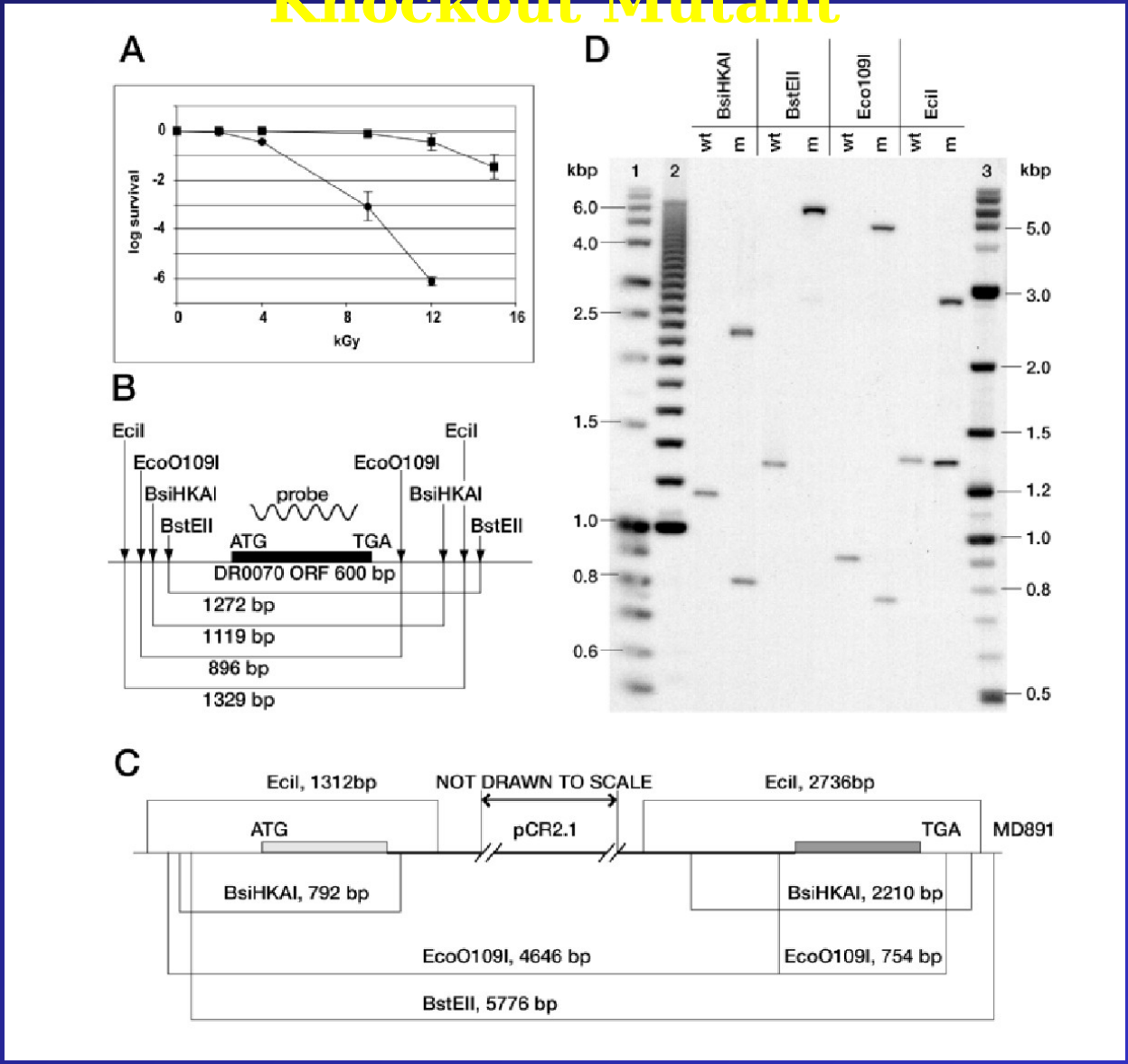
Coordinated Expression of Genes Belonging to the Same Operon or Multi-Subunit Complex



Expression Patterns of Predicted Transcriptional Regulators Potentially Involved in the Regulation of *D. radiodurans'* Irradiation-Response

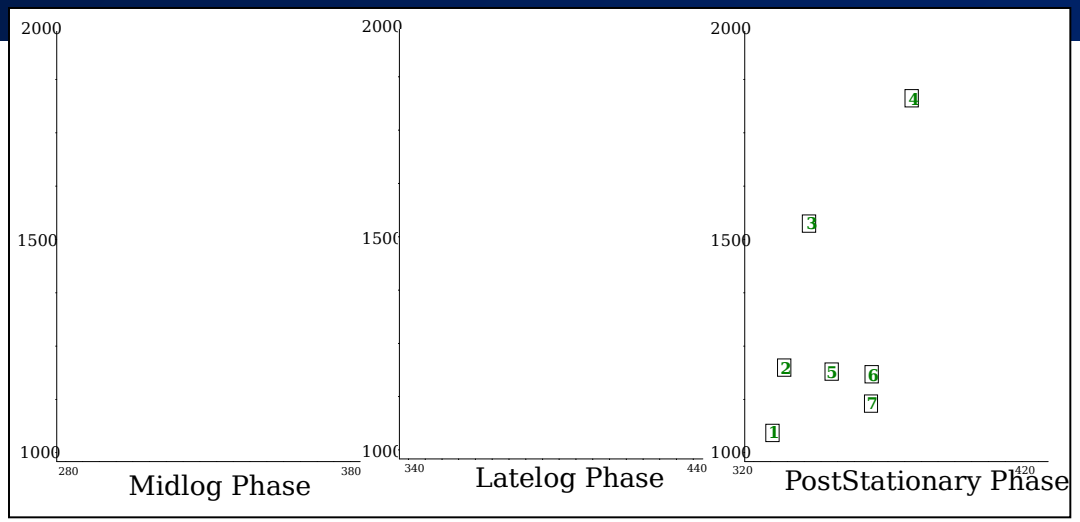


Resistance Phenotype, Construction, and Genomic Structure for the DR0070 (*ddrB*) Knockout Mutant



Different Functional Groups of Genes and Operons of *D. radiodurans* with Early-Mid Expression *recA*-like Patterns and the Result of ORF Disruption (marked in green)

	Gene_ID[a]	Fun-ction group [b]	Protein description and comments	Maximum induction level (fold)	Maximum induction time (hr)	Result of ORF disruption
1	DR0003	-	Uncharacterized protein	14	1.5	m resist to 9 kGy
2	DR0050	L	DinB/YfiT family protein	4	3	
3	DR0051	S	Small cysteine-rich protein of the HesB family	6	3	
4	DR0052	-	Uncharacterized conserved protein	7	1.5	
5	DR0053	L	DinB/YfiT family protein	10	3	m resist to 9 kGy
6	DR0070	ST?	Uncharacterized protein	4	9	m sens to 5 kGy, homozyg
7	DR0103	R	Predicted metal-binding protein	6	1.5	
8	DR0105	ST	LEA76 family desiccation resistance protein	3	0.5	m resist to 9 kGy
9	DR0140	-	Uncharacterized protein	6	1.5	m resist to 9 kGy
10	DR0160	-	Conserved membrane protein	4	1.5	
11	DR0161	S	AmsJ/WcaK related protein, possibly involved in exopolysaccharide biosynthesis	8	1.5	
12	DR0203	R	Uncharacterized membrane protein	4	1.5	
13	DR0204	R	Uncharacterized membrane protein	6	3	



Molecular Weight

4325
3850
3375
2900
2425
1950
1475
1000

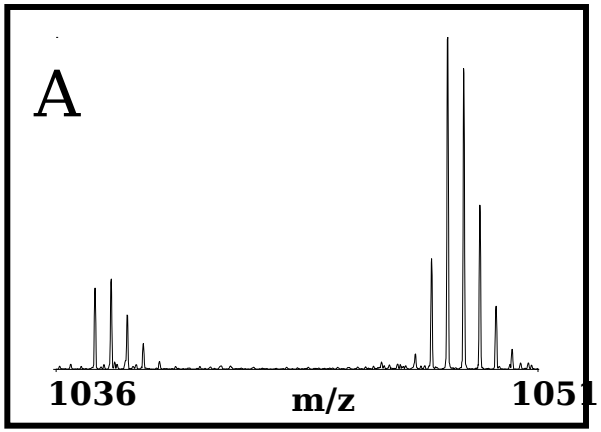
100 200 300 400 500 600 700 800

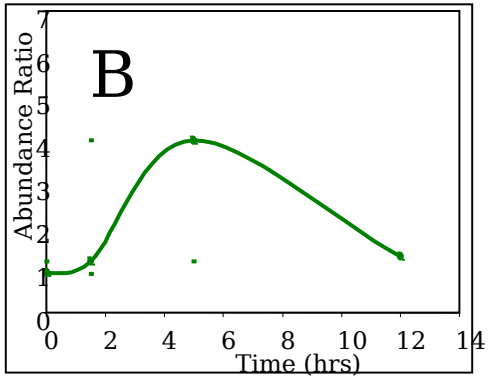
Spectrum number
(Time ⟶)

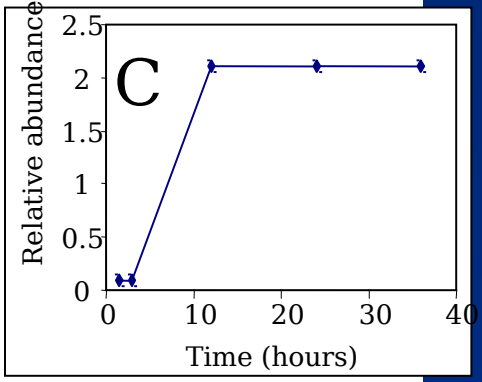
See next slide for comm

2D display of a capillary LC-FTICR analysis in which >50,000 putative polypeptides were detected form a tryptic digest of proteins isolated from D. radiodurans harvested in mid log phase. The inset shows three 2-D displays for peptides isolated from D. radiodurans harvested in midlog, late log and post stationary phases. Within the inset, spot size has been adjusted to show highly abundant species as larger spots creating a display similar to that of a 2-D PAGE.

Less abundant — More abundant





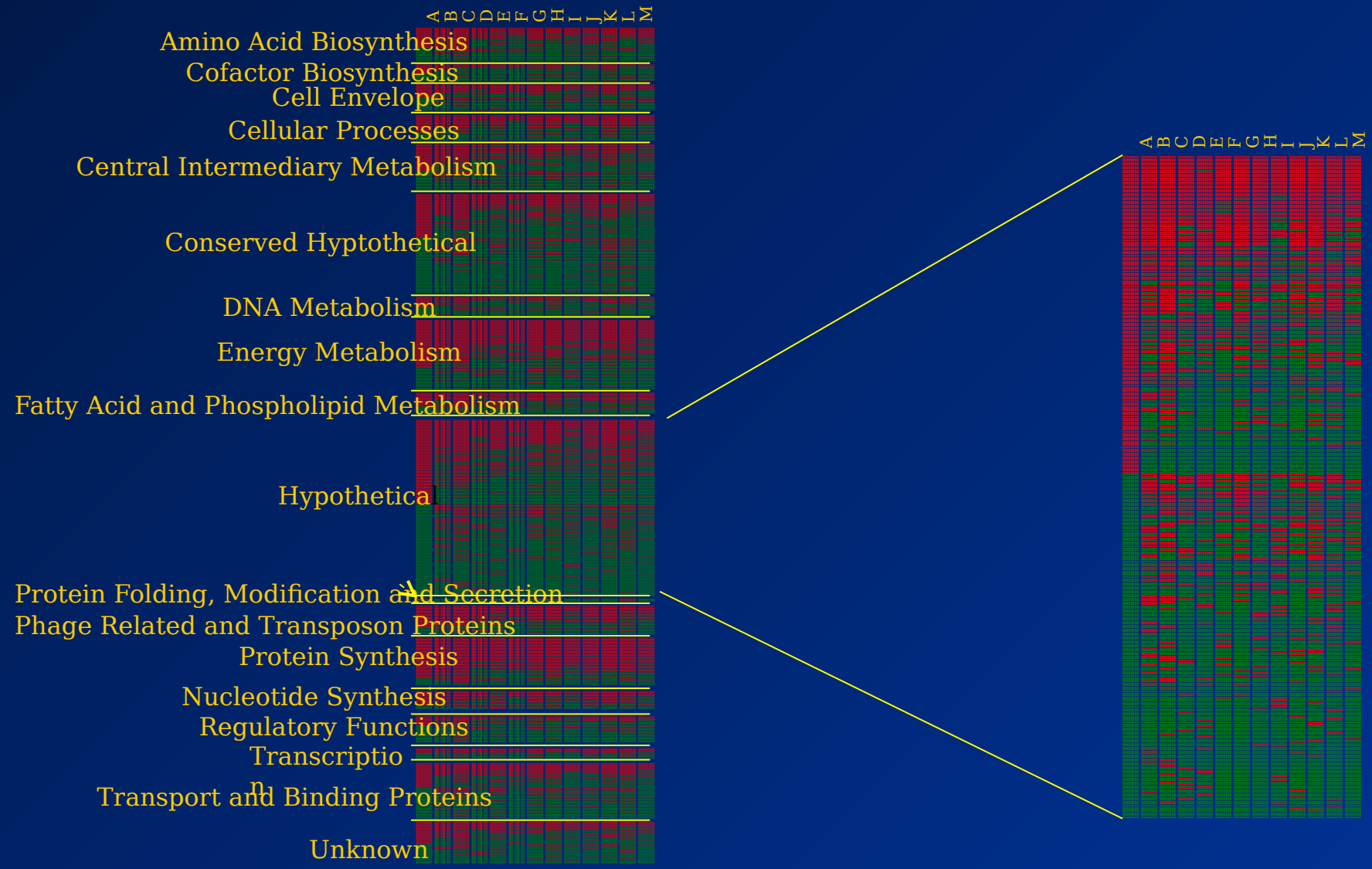


Molecular Weight

3000
2000
1000

150
300
450
600

Spectrum number
(Time →)


A B C D E F G H I J K L M
Amino Acid Biosynthesis
Cofactor Biosynthesis
Cell Envelope
Cellular Processes
Central Intermediary Metabolism
Conserved Hyptothetical
DNA Metabolism
Energy Metabolism
Fatty Acid and Phospholipid Metabolism
Hypothetical
Protein Folding, Modification and Secretion
Phage Related and Transposon Proteins
Protein Synthesis
Nucleotide Synthesis
Regulatory Functions
Transcriptio
n
Transport and Binding Proteins
Unknown
A B C D E F G H I J K L M


The End